3-Monochloropropane-1,2-diol (3-MCPD)

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Occurrence and Use

- Used as a dye intermediate, solvent for cellulose acetate, and to lower the freezing point of dynamite
- Registered as a restricted use rodenticide
- Formed in foods during processing, cooking and storage
- Found in a variety of foods containing acid-hydrolyzed vegetable protein (HVP) and some foods without acid-HVP
- Detected in packaging materials and drinking water

Carcinogenicity Studies in Animals

- Studies in mice:
  - Topical application in Swiss mice (1)
  - Subcutaneous injection (s.c.) in Swiss mice (1)
  - Drinking water studies – B6C3F_1 mice (2)
- Studies in rats:
  - Oral gavage studies – Sprague-Dawley rats (2)
  - Drinking water studies – Fischer rats (2); Sprague-Dawley rats (2)

19-Month Studies in Mice
(Van Durren *et al*., 1974)

- Dermal study (2 mg, 3/week) in female Swiss mice (n = 50)
  - No treatment-related skin tumors

- S.C. injection study (1 mg, 1/week) in female Swiss mice (n = 50)
  - No treatment-related neoplastic findings

104-Week Studies in Mice
(Jeong *et al*., 2010)

- Male and female $B6C3F_1$ mice (n = 50)
- Drinking water (0, 30, 100, 300/200 ppm)
  - High-dose mice had significantly decreased body weight, food and water consumption
  - No treatment-related neoplastic findings

Gavage Studies in Rats
(Weisburger *et al*., 1981)

- Male and female Sprague-Dawley rats (treated: n = 26; controls: n = 20)
- 30/35 or 60/70 mg/kg$_{bw}$, 2/week for 72 weeks; observed for additional 32 weeks
  - No treatment-related neoplastic findings

Drinking water studies in Fischer rats

(Sunahara *et al*., 1993)

Organ and Lesion	3-MCPD Concentration (ppm)				Trend test p-value
	0	20	100	500	
Male: Testis					
Leydig cell adenoma	38/50	43/50	50/50***	47/50*	≤0.01
Leydig cell carcinoma	0/50	0/50	0/50	3/50	≤0.001
Male: Mammary gland					
Fibroadenoma	0/45	0/48	2/47	10/49***	≤0.0001
Adenoma	0/45	0/48	1/47	1/49	NS
Adenocarcinoma	0/45	0/48	1/47	1/49	NS

Drinking water studies in Fischer rats
(Sunahara *et al*., 1993) (continued)

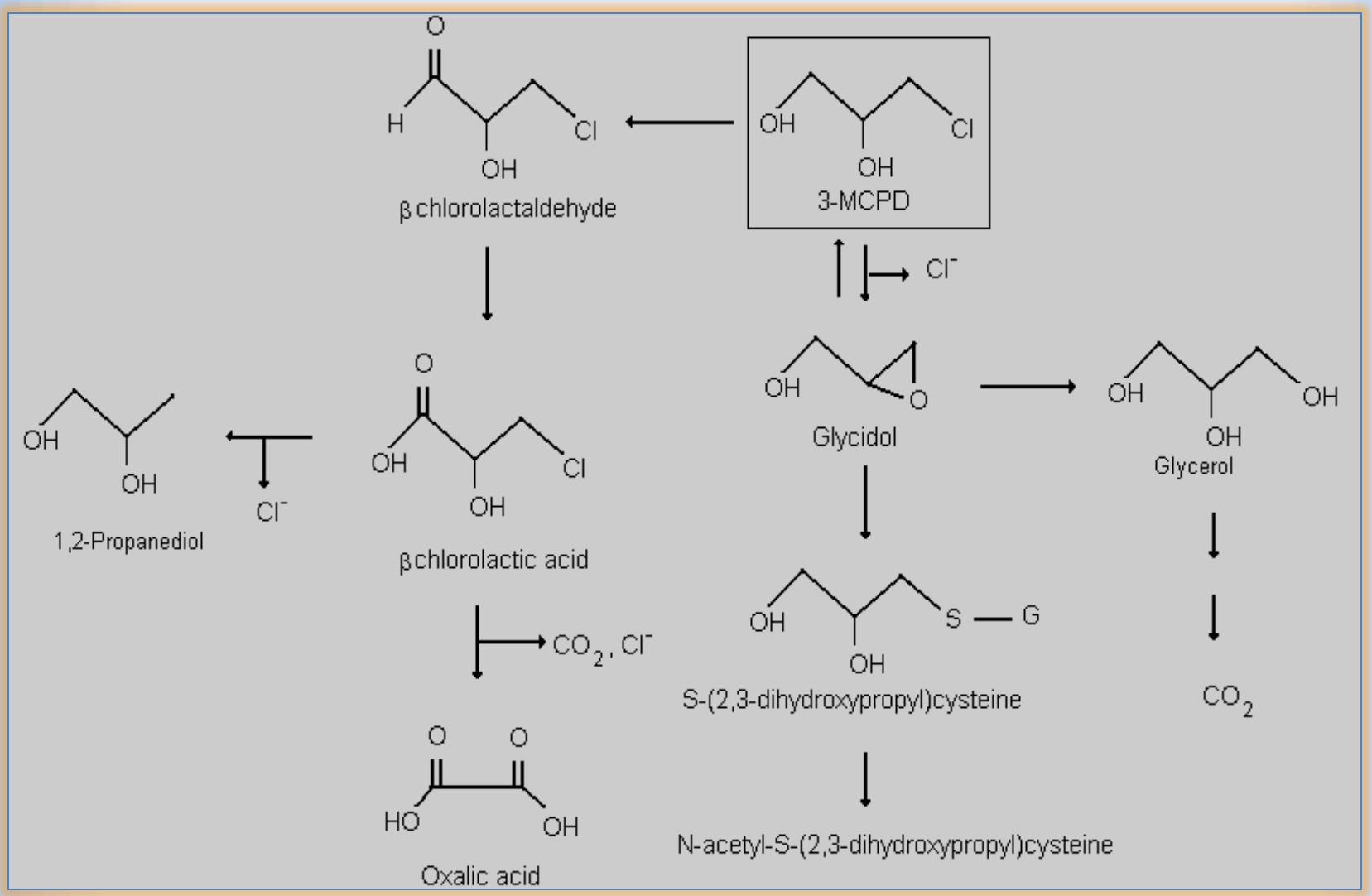
Organ and Lesion	3-MCPD Concentration (ppm)				Trend test p-value
	0	20	100	500	
Male: Kidney					
Tubular adenoma	0/50	0/50	1/50	5/50*	≤0.01
Female: Kidney					
Tubular adenoma	0/50	1/50	0/50	9/50**	≤0.0001

Drinking water studies in S-D rats

(Cho *et al*., 2008)

Organ and Lesion	3-MCPD concentration (ppm)				Trend test p-values
	0	25	100	400	
Male: Testis Leydig cell tumors	1/50	1/50	4/50	14/50****	≤ 0.001
Male: Kidney					
Tubular adenoma	0/50	0/50	1/50	4/50	≤ 0.01
Tubular carcinoma	0/50	0/50	0/50	5/50**	≤ 0.001
Tubular adenoma and carcinoma (combined)	0/50	0/50	1/50	7/50***	≤ 0.0001
Female: Kidney					
Tubular adenoma	0/50	0/50	1/50	6/50**	≤0.001
Tubular carcinoma	1/50	0/50	1/50	3/50	=0.06
Tubular adenoma and carcinoma (combined)	1/50	0/50	2/50	9/50***	≤0.0001

Proposed Metabolic Pathways for 3-MCPD



In vitro Genotoxicity

- Positive in
  - *Salmonella typhimurium* reverse mutation assay
    - TA 1535 and TA 100 (+/- S9) base-pair substitution
    - TA 98 (-S9) frameshift mutation
  - *S. plombe* forward mutating assay (-S9)
  - Gene mutation in mouse lymphoma cells (+S9)
  - SCEs in Chinese hamster V79 cells (+/- S9)
  - DNA damage in CHO cells (-S9)
- Negative in
  - *E. coli* (TN930, TN1080 and WP2) (+/- S9)
  - DNA synthesis inhibition assay in HeLa cells (+/- S9)

In vivo Genotoxicity

- Negative in various *in vivo* assays
  - *Drosophila* somatic mutation assay (Wing spot)
  - Dominant lethal mutation assay in male mice
  - Bone marrow micronucleus assay in rats and mice
  - Unscheduled DNA synthesis in liver cells of male rats
  - DNA damage in various tissues and blood cells of male rats

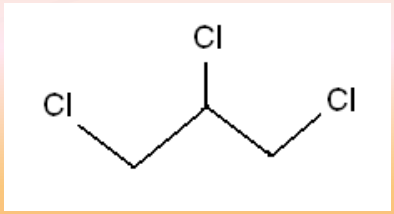
Immune System Effects

- ***In Vivo* (mice)**
  - ↓ absolute and relative thymus weights
  - ↓ natural killer cell activity
  - ↓ antibody production to sheep red blood cells
  - ➢ *Suggests reduced capacity for cell lysis and tumor surveillance*
- ***In Vitro* (mice)**
  - ↓ proliferative response to con A, anti-CD3 antibody, and lipopolysaccharides
  - ↓ spleen cell production of IFN-gamma, IL-10, and IL-4
  - ↓ macrophage production of TNF-alpha and NO (at non-cytotoxic doses)
  - ➢ *Suggests impaired T & B cell function, altered regulation of inflammatory response*

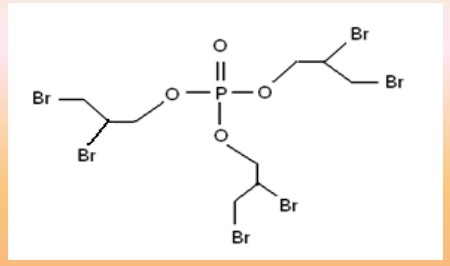
MALIGNANT TRANSFORMATION OF MOUSE M2 FIBROBLASTS

	3-MCPD (µg/mL)					
	Control	100	250	500	100	2000
Number of transformed foci/number of treated dishes	0/24	2/27	10/25 p<0.0001	14/27 p<0.0001	4/26	0/22

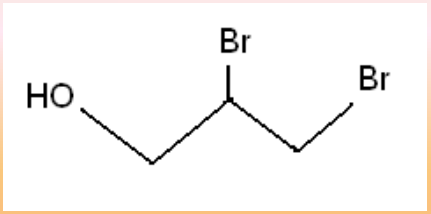
Structure activity comparison



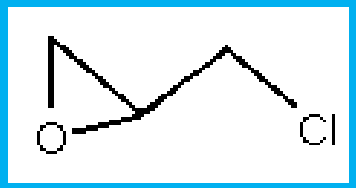
1,2,3-Trichloropropane



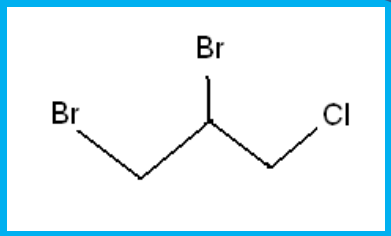
Tris (2,3,-dibromopropylphosphate)



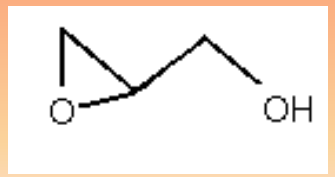
2,3-Dibromo-1-propanol



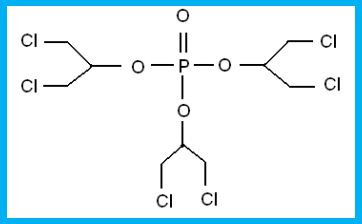
Epichlorohydrin



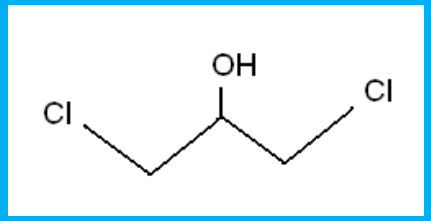
Dibromochloropropane (DBCP)



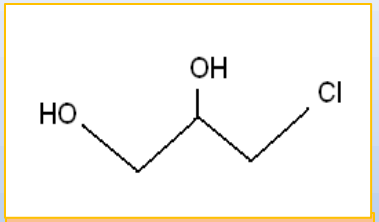
Glycidol



Tris (1,3-dichloro-2-propyl) phosphate



1,3-Dichloro-2-propanol



3-MCPD

Possible Mechanisms of Action

- Genotoxicity
- Inhibition of glycolysis
  - Glyceraldehyde-3-phosphate dehydrogenase (GAPDH) inhibition
    - Regulation of DNA repair, cell death, cell cycle progression, mRNA stability
- Kidney toxicity
- Immune effects
- Hormonal effects

Summary of Evidence

❑ Animal evidence for carcinogenicity

- ➢ Tumors in both sexes of two strains of rats
- ➢ Tumors at multiple sites in two strains of ♂ rats

Kidney tumors

- S-D rats: ↑ combined adenoma & carcinoma in ♀ & ♂
  - Rare
  - Early onset in ♂
- Fischer rats: ↑ adenoma in ♀ & ♂

Mammary tumors

- ♂ Fischer rats: ↑ fibroadenoma; adenoma and adenocarcinoma observed in mid- & high-dose
  - Uncommon

Leydig cell tumors

- Fischer rats: ↑ combined adenoma & carcinoma
- S-D rats: ↑ in Leydig cell 'tumors'

Summary of Evidence (continued)

- *In vitro* genotoxicity in a variety of systems
- Malignant transformation of cells
- Metabolism to glycidol, a genotoxic carcinogen
- Structurally similar to six carcinogens